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USPT	end near0 label\$ near0 ribonucl\$	3	<u>L3</u>
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USPT	fluorescen\$ same label\$ same ribonucleotide	47	<u>L1</u>

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L3: Entry 2 of 3

File: USPT

Aug 12, 1997

DOCUMENT-IDENTIFIER: US 5656742 A

TITLE: Ring-opened purine compounds for labeling polynucleotides

DEPR:

The reaction is incubated at 37.degree. C. for 30 minutes and terminated by adding 2 .mu.l of 0.5M EDTA. End-labeled oligonucleotide is separated from unincorporated Bio-dGTP by centrifugation through a BioGel P2 spin column. If desired, TdT is removed by phenol extraction and the oligonucleotide is precipitated from the aqueous phase by the addition of two volumes of absolute ethanol and incubation at -70.degree. C. for at least 1 hour. The use of CoCl.sub.2 allows end-labeling of ribonucleotides as well as deoxyribonucleotides.

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Term: 11 same (advantag\$ or useful\$)

Display: 10 **Documents in Display Format:** CIT **Starting with Number** 1

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USPT	fluorescen\$ same label\$ same ribonucleotide	47	<u>L1</u>

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L6: Entry 1 of 14

File: USPT

Aug 7, 2001

DOCUMENT-IDENTIFIER: US 6270976 B1

TITLE: Characterizing nucleic acid by mass spectrometry

DEPR:

This approach is necessary if RNA polymerases are used in conjunction with ribonucleotides or their analogues since most RNA polymerases use promoter sequences rather than primers and so incorporation of labels would have to be effected via labelled nucleotides.

DEPR:

One possible fragmentation resistant DNA 'analogue' that already has appropriate polymerases is of course RNA. RNA is chemically less stable than DNA but is more resistant to fragmentation in the mass spectrometer. Generally RNA is disliked as a material to work with as it is so easy to contaminate with degrading enzymes in manual experiments. However for automated high throughput sequencing this may not be a significant problem as contamination by RNases, etc. can be much more rigorously controlled. For use in sequencing one would require terminating ribonucleotides or analogues that are accepted by an RNA polymerase. Such terminators could be generated by synthesising ribonucleotides with the 3' hydroxyl blocked. The blocking group could be a linker to a cleavable mass label identifying the nucleotide.